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**Effects of carbohydrate dose and frequency on metabolism, gastrointestinal discomfort  
and cross-country skiing performance**

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**Running Head:** Carbohydrate ingestion and cross-country skiing

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There are no conflicts of interest to declare. No external funding was received for this study.

## Abstract

This study investigated carbohydrate ingestion of varied doses and frequencies during a simulated cross-country skiing time-trial. Ten men and three women (age:  $30 \pm 7$  y;  $\dot{V}O_{2\max}$ :  $59.6 \pm 5.7$  mL·kg<sup>-1</sup>·min<sup>-1</sup>) completed four, 30-km classic technique roller-skiing time-trials on a treadmill. A 1:1 maltodextrin-fructose carbohydrate solution was provided at high (2.4 g·min<sup>-1</sup>; HC) and moderate (1.2 g·min<sup>-1</sup>; MC) ingestion rates, each at high (six feeds; HF) and low (two feeds; LF) frequencies. In the LF trials blood glucose was elevated following carbohydrate ingestion (at 4 and 19 km) but was reduced at 14 and 29 km compared to HF strategies ( $p \leq 0.05$ ). Gastrointestinal discomfort was higher in HC-LF compared with all other trials ( $p \leq 0.05$ ). Whole-body lipid oxidation was lower and carbohydrate oxidation was higher in LF compared with HF trials ( $p \leq 0.05$ ). While performance time was not significantly different between trials (140:11 $\pm$ 15:31, 140:43 $\pm$ 17:40, 139:12 $\pm$ 15:32 and 140:33 $\pm$ 17:46 min:s in HC-HF, HC-LF, MC-HF and MC-LF, respectively;  $p > 0.05$ ), it was improved with trial order ( $p < 0.001$ ). There was no effect of order on any other variable ( $p > 0.05$ ). Altering carbohydrate dose or frequency does not affect cross-country ski performance. However, low frequency carbohydrate ingestion resulted in poorer maintenance of euglycaemia, reduced lipid oxidation and increased gastrointestinal discomfort.

**Key words:** endurance performance, glucose, multiple-transportable carbohydrate, Nordic skiing, roller-skiing

## Introduction

Carbohydrate ingestion during endurance exercise lasting longer than one hour improves endurance capacity and performance (Jeukendrup, 2014). However, recommendations to ingest 0.5-1.0 g·min<sup>-1</sup> of carbohydrate during exercise lasting up to three hours (Rodriguez et al., 2009; Jeukendrup, 2014) may be somewhat conservative. Specifically, maximal exogenous carbohydrate oxidation rates of ~ 1.0-1.1 g·min<sup>-1</sup> appear to peak at ingestion rates of ~ 1.0-1.2 g·min<sup>-1</sup> when using single-transportable carbohydrates (Wagenmakers et al., 1993). Furthermore, when carbohydrate is ingested at higher rates in the form of multiple-transportable carbohydrates (i.e., glucose and fructose mixtures), which utilise different intestinal transporter molecules, oxidation rates can increase (Jentjens et al., 2004; Jentjens et al., 2004; Jentjens & Jeukendrup, 2005). For example, exogenous carbohydrate oxidation rates as high as 1.75 g·min<sup>-1</sup> have been reported when carbohydrate is ingested at 2.4 g·min<sup>-1</sup> as a 24 % solution of 1:1 glucose-fructose (Jentjens & Jeukendrup, 2005).

Several studies have applied the theoretical principle of ingesting high rates (e.g., 1.4-2.4 g·min<sup>-1</sup>) of multiple-transportable carbohydrates and demonstrated performance benefits when compared with isoenergetic doses of single carbohydrates and/or a placebo (Currell & Jeukendrup, 2008; Triplett et al., 2010; McGawley et al., 2012; Rowlands et al., 2012). However, when ingestion of 1.55 g·min<sup>-1</sup> of a glucose-fructose mixture was compared to ingestion of glucose-only at a rate approximating the recommended dose (1.03 g·min<sup>-1</sup>) no additional ergogenic effect was demonstrated (Baur et al., 2014). Moreover, two studies investigating the dose-response relationship between multiple-transportable carbohydrate ingestion and performance suggest that cycling performance may peak at an ingestion rate of ~ 1.2-1.3 g·min<sup>-1</sup> (Mitchell et al., 1989; Smith et al., 2013), which differs from the previously

70 stated 2.4 g·min<sup>-1</sup> ingestion rate that elicits the highest reported exogenous carbohydrate  
71 oxidation rate (Jentjens & Jeukendrup, 2005).

72  
73 While studies investigating the ergogenic effect of carbohydrate have been undertaken in  
74 cycling and running (Mitchell et al., 1989; Tsintzas et al., 1996; Currell & Jeukendrup, 2008;  
75 Triplett et al., 2010; Smith et al., 2013; Baur et al., 2014), the effects of carbohydrate  
76 ingestion during cross-country skiing are less well known. Due to the whole-body nature of  
77 cross-country skiing (Björklund et al., 2010), glycogen depletion is evident in both the upper  
78 and lower body (Bergström et al., 1973). Moreover, the double-poling skiing technique results  
79 in greater carbohydrate utilisation, measured by the respiratory exchange ratio, compared to  
80 uphill running at a matched relative intensity (Losnegard et al., 2014). Thus the carbohydrate  
81 requirement during cross-country skiing may be different from the optimal dose of 1.0-1.3  
82 g·min<sup>-1</sup> identified for cycling performance (Smith et al., 2013). The only study to have  
83 assessed the ergogenic effect of carbohydrate ingestion during cross-country skiing found a  
84 non-significant trend for improved 50-km race performance with a 67 % glucose polymer  
85 syrup providing ~ 9 g·min<sup>-1</sup> of carbohydrate compared to an equal volume of a 2.5 % glucose  
86 solution providing ~ 0.3 g·min<sup>-1</sup> of carbohydrate (Viinamaki et al., 1989). However, it is  
87 difficult to draw practical conclusions from that study as the glucose polymer syrup provided  
88 glucose at a much higher rate than can be absorbed from the gut and oxidised (Wagenmakers  
89 et al., 1993).

90  
91 Compared with absolute dose, the timing and distribution of carbohydrate ingestion during  
92 endurance performance have received limited research attention. Mechanistically, the  
93 frequency of carbohydrate feeding appears to have little effect on the peak exogenous  
94 carbohydrate oxidation rates or the time taken to reach this peak (Jeukendrup & Jentjens,

2000). However, performance is the combination of exogenous carbohydrate oxidation rates and other positive and negative factors such as gastrointestinal (GI) discomfort, which require consideration when providing practical recommendations. Only one study has compared the effects of different carbohydrate ingestion frequencies on performance (McConnell et al., 1996). In that study, the isoenergetic intake of three highly concentrated boluses of carbohydrate after 90, 105 and 120 minutes of steady-state cycling did not improve time-trial (TT) performance compared with a placebo. However, due to differences in the timing of ingestion (i.e., late in exercise vs. throughout exercise) in this study, the idea that more frequent carbohydrate ingestion versus fewer large boluses may be beneficial to performance is still to be demonstrated. In addition, the effects of feeding frequency using multiple-transportable carbohydrates have not been assessed.

The aims of the current study were to investigate whether, during cross-country skiing, there is a greater ergogenic effect of multiple-transportable carbohydrate ingestion (i) at a high rate (i.e.,  $2.4 \text{ g} \cdot \text{min}^{-1}$ ) compared to a moderate rate (i.e.,  $1.2 \text{ g} \cdot \text{min}^{-1}$ ) and (ii) as small and frequent feeds (i.e., six separate feeds) compared to large and infrequent feeds (i.e., two separate feeds). The final aim was (iii) to characterise the GI discomfort and metabolic responses to each carbohydrate ingestion strategy, which are of practical and mechanistic importance to the recommendation of carbohydrate ingestion strategies. It was hypothesised that (i) cross-country ski performance would be improved with the ingestion of  $2.4 \text{ g} \cdot \text{min}^{-1}$  compared to  $1.2 \text{ g} \cdot \text{min}^{-1}$  of a 1:1 maltodextrin-fructose solution, and (ii) that cross-country ski performance would be improved with small and frequent feeds compared to large and infrequent feeds.

## Materials and Methods

## Participants

Thirteen well-trained cross-country skiers including ten men and three women (mean  $\pm$  SD: age,  $30 \pm 7$  years; body mass,  $74.1 \pm 8.7$  kg; body fat,  $17.2 \pm 3.8$  %; maximal oxygen uptake ( $\dot{V}O_{2\max}$ ),  $59.6 \pm 5.7$  mL $\cdot$ kg $^{-1}\cdot$ min $^{-1}$ ) were recruited to participate. All participants had previously competed in at least two **classic technique** cross-country ski races of 30 km or longer (mean  $\pm$  SD races  $\geq 30$  km:  $21 \pm 7$ ) and were partaking in regular cross-country ski training (mean  $\pm$  SD training per week:  $9 \pm 5$  h) **and local races. The participants were experienced in classical skiing but were not specialists or competing at an international level.** They were fully informed of the study procedures and their right to withdraw before providing written consent to participate. The study was pre-approved by the Regional Ethical Review Board, Umeå University, Umeå, Sweden.

## Experimental Overview

Participants attended five laboratory-based testing sessions conducted at the same time of day ( $\pm 2$  h), each separated by at least four days. Prior to the testing period participants were assessed for body composition via dual-energy x-ray absorptiometry (DXA). The first laboratory testing session was used to assess  $\dot{V}O_{2\max}$  and familiarise participants with the experimental procedures. The four subsequent sessions involved a 30-km **classic technique** roller-skiing TT on a treadmill using the following four carbohydrate ingestion strategies, which were provided in a randomised, counter-balanced order: high carbohydrate – high frequency (HC-HF), high carbohydrate – low frequency (HC-LF), moderate carbohydrate – high frequency (MC-HF) and moderate carbohydrate – low frequency (MC-LF). **Carbohydrate ingestion rates were provided in a double-blind manner.** The high and moderate carbohydrate beverages were designed to provide ingestion rates of 2.4 g $\cdot$ min $^{-1}$  (HC) and 1.2 g $\cdot$ min $^{-1}$  (MC) provided as 24 % and 12 % solutions, respectively, with all

solutions consisting of a 1:1 maltodextrin-fructose mixture flavoured with a non-caloric tropical fruits-flavoured sweetener (Table 1). **Participants consumed the first feed five minutes prior to each trial and then at 5-km intervals in the high frequency (HF) trials (i.e., six feeds in total) or after 15 km in the low frequency (LF) trials (i.e., two feeds in total).** Participants were given 30 and 90 s to consume each feed in the HF and LF trials, respectively, so that total drinking time was matched. Drinking times during the 30-km TTs were, however, designed to be lower in the LF trials (90 s) compared with the HF trials (150 s).

**\*\* INSERT TABLE 1 AROUND HERE \*\***

## **Equipment**

All tests were performed using the same pair of pre-warmed roller-skis (Pro-Ski Classic C2, Sterners, Dala-Järna, Sweden) to minimise variations in rolling resistance (Ainegren et al., 2008). The  $\dot{V}O_{2\max}$  test and the 30-km TTs were completed on a motor-driven treadmill (Rodby RL 3000, Rodby, Vänge, Sweden) with lasers that automatically increased or decreased the velocity if the athlete moved to the front or rear of the belt, respectively, maintaining a constant velocity otherwise. Respiratory variables were measured during the  $\dot{V}O_{2\max}$  test and the 30-km TTs using a mixed expired-air procedure with an ergospirometry system (AMIS 2001 model C, Innovision A/S, Odense, Denmark) equipped with an inspiratory flow-meter. The gas analysers were calibrated with a high-precision mixture of 16.0 %  $O_2$  and 4.5 %  $CO_2$  (Air Liquide, Kungsängen, Sweden) and the flow-meter was calibrated at three rates with a 3-L air syringe (Hans Rudolph, Kansas City, USA). Heart rate was monitored continuously (RS400, Polar Electro Oy, Kempele, Finland) and blood lactate and glucose concentrations were measured from fingertip samples (Biosen C-line, EKF



diagnostic GmbH, Magdeburg, Germany). All blood samples were mixed on a rocker and placed on ice until analysis at the completion of each testing session. Rating of perceived exertion (RPE) was assessed using the Borg scale (Borg, 1973) and GI discomfort was assessed using a modified Borg scale, where scores of six and 20 represent no discomfort and maximum discomfort, respectively (McGawley et al., 2012).

## **Familiarisation**

Prior to the start of the study, participants undertook supervised treadmill roller-skiing training sessions where a range of velocities, gradients and skiing techniques were practiced in each session. These sessions were continued until the participant was comfortable with treadmill roller-skiing and controlling the automatic velocity adjustment on the treadmill. The final supervised session included a familiarisation to the  $\dot{V}O_{2\max}$  test protocol and the 5-km lap used during the 30-km TTs.

Formal familiarisation to the 30-km TT protocol was provided upon completion of the  $\dot{V}O_{2\max}$  test. **After 15-20 min of rest**, participants completed the first 15 km of the 30-km TT course using the HF protocol. The trial included familiarisation with expired gas collection, the subjective rating scales and the drinking procedures used during the 30-km TTs. Participants received carbohydrate at a rate of  $1.8 \text{ g} \cdot \text{min}^{-1}$ , which was designed to be an average of the MC and HC trials, and were instructed to pace themselves as if they were completing the whole 30-km course. The time taken to complete the 15-km familiarisation was recorded and used to calculate the pre-trial feed volume in the subsequent data collection trial (see equations and explanation in Supporting Information).

## **Testing**

*Body composition*

After measuring height and body mass (Seca 764, Hamburg, Germany), body composition was measured using the Lunar iDXA (GE Healthcare, Waukesha, WI, USA). Participants were tested in a fasted and rested state between 06:00 and 09:00 (Nana et al., 2012).

$\dot{V}O_{2max}$

$\dot{V}O_{2max}$  was assessed using a 600-m diagonal-stride TT protocol at 7°, which has been shown to produce similar  $\dot{V}O_{2max}$  values to a traditional incremental ramp test (McGawley & Holmberg, 2014). The protocol was performed as previously described, with the exception of the velocity during the first 100 m being fixed at 8.8, 9.8 or 10.8 km·h<sup>-1</sup>, depending on the skiing ability of the participant.

*30-km time trials*

Prior to testing participants were instructed to abstain from alcohol for at least 24 h and from caffeine prior to testing on the day of each trial. Participants arrived at the laboratory having consumed a pre-race meal of their choice two hours prior to arrival and were asked to record a food and exercise diary for the day prior to and the day of the first 30-km trial so that activity and food intake could be replicated prior to each subsequent trial. Upon arrival at the laboratory, body mass was measured in minimal clothing and a resting blood sample, a baseline GI discomfort score and a two-minute resting expired-air sample were collected. Participants then undertook a 10-minute self-paced warm-up, which was recorded and replicated prior to each subsequent trial. Two minutes after completion of the warm-up, participants received their pre-trial feed. The participant was then fitted with a mouthpiece, nose-clip and roller-skis. The 30-km self-paced **classic technique** TT began five minutes after the participant began consuming the first feed. The TT consisted of six laps of five kilometres

(Figure 1). Figure 1A outlines when blood samples, expired-air samples and scores of GI discomfort and RPE were collected, and when carbohydrate feedings were received throughout each trial. The treadmill was slowed to a fixed velocity of 4.5 km·h<sup>-1</sup> at 4° for 80 s during the blood sampling periods and for 30 and 90 s during drinking periods for the HF and LF trials, respectively. Mean heart rate,  $\dot{V}O_2$  and  $\dot{V}CO_2$  were calculated on each lap during the final minute of one section at 1° (designed to induce the double-poling technique) and one section at 7° (designed to induce the diagonal-stride technique), as displayed in Figure 1B. Total carbohydrate and lipid oxidation rates were calculated for the 1° sections from the  $\dot{V}O_2$  and  $\dot{V}CO_2$  using the high-intensity equations of Jeukendrup and Wallis (Jeukendrup & Wallis, 2005):

$$\text{Carbohydrate oxidation rate} = 4.210 \times \dot{V}CO_2 - 2.962 \times \dot{V}O_2$$

$$\text{Lipid oxidation rate} = 1.695 \times \dot{V}O_2 - 1.701 \times \dot{V}CO_2$$

Total carbohydrate and lipid oxidation rates were not calculated during the 7° sections as  $\dot{V}O_2$  exceeded 80 % of  $\dot{V}O_{2\max}$ , rendering the stoichiometric equations invalid (Jeukendrup & Wallis, 2005). Time and velocity were blinded from the participants throughout all trials and standardised verbal feedback and encouragement to complete the TT as fast as possible were provided continuously by the experimenter. Time to complete the 30-km TT was recorded.

**\*\* INSERT FIGURE 1 AROUND HERE \*\***

#### *Feed volume calculations and ingestion rates*

Due to the complications of providing carbohydrate as a rate (in g·min<sup>-1</sup>) during a TT when feeds are provided at set distances and time is previously unknown, several equations were developed to calculate the required volumes throughout the TTs. Equations and explanation of calculations can be found online in the Supporting Information. In order to maintain

double-blinding, pre-trial volumes and required fluid ingestion rates were calculated and beverages were made by a researcher external to the project.

### *Statistical analysis*

The Statistical Package for the Social Sciences (SPSS) 18.0 was used to carry out statistical analyses. Data are expressed as mean  $\pm$  standard deviation. Mean values for feed volume, blood lactate, heart rate, RPE,  $\dot{V}O_2$  and whole-body carbohydrate and lipid oxidation were calculated from six data points throughout the TT, one from each 5-km lap. Mean values for GI discomfort were calculated from 12 values, two from each 5-km lap. Order effect and differences in total ingested fluid volumes were assessed using one-way repeated measures ANOVAs. Differences in performance time, carbohydrate ingestion rate, feed volume, blood lactate concentration, GI discomfort, heart rate, RPE,  $\dot{V}O_2$  and whole-body carbohydrate and lipid oxidation rates were assessed by two-way repeated measures ANOVAs, with carbohydrate ingestion rate and frequency entered as separate factors. Distance-series data of blood glucose concentration and GI discomfort were analysed by two-way repeated measures ANOVAs, with treatment and distance entered as separate factors. Multiple comparisons were corrected by a Ryan-Holm-Bonferroni stepwise adjustment to prevent inflation of the type I error rate (Ludbrook, 1998). The level of significance was set at  $p \leq 0.05$ .

## **Results**

### *Carbohydrate ingestion*

The rates of carbohydrate ingestion were  $2.36 \pm 0.04$  and  $2.34 \pm 0.04$  g $\cdot$ min $^{-1}$  in the HC-HF and HC-LF trials, respectively, and  $1.18 \pm 0.03$  and  $1.18 \pm 0.02$  g $\cdot$ min $^{-1}$  in the MC-HF and MC-LF trials, respectively. The ingestion rate in the HC trials was significantly higher than in

the LC trials ( $p < 0.001$ ), but was not significantly different between frequencies ( $p = 0.099$ ). Total ingested fluid volumes were  $1380 \pm 145$ ,  $1371 \pm 167$ ,  $1369 \pm 136$  and  $1381 \pm 160$  ml in the HC-HF, HC-LF, MC-HF and MC-LF trials, respectively, and did not significantly differ between trials ( $p = 0.714$ ). These feeds were provided in feed volumes of  $230 \pm 24$  and  $228 \pm 23$  in the HC-HF and MC-HF trials, respectively, and  $686 \pm 83$  and  $690 \pm 80$  ml in the HC-LF and MC-LF trials, respectively. The feed volumes in the LF trials was significantly higher than in the HF trials ( $p < 0.001$ ), but was not significantly different between ingestion rates ( $p = 0.667$ ).

#### *Blood metabolites*

In LF trials, poorer maintenance of euglycaemia was apparent compared to HF trials. There was a significant treatment\*distance interaction effect ( $p = 0.020$ ), but no significant effect of treatment ( $p = 0.110$ ) or distance ( $p = 0.120$ ) for blood glucose concentration (Figure 2). Post-hoc analyses revealed elevated blood glucose concentrations in both LF trials compared to HF trials in the blood sample following the first carbohydrate bolus (i.e., at 4 km; HC:  $p = 0.041$ , MC:  $p = 0.002$ ) and a tendency for an increased blood glucose concentration after the second carbohydrate bolus (i.e., at 19 km; HC:  $p = 0.125$ , MC:  $p = 0.079$ ). Blood glucose concentrations were significantly lower in the LF compared with the HF trials at 14 km (HC:  $p = 0.046$ , MC:  $p = 0.004$ ) and at 29 km (HC:  $p = 0.046$ , MC:  $p = 0.047$ ). There was no effect of carbohydrate ingestion rate, frequency or carbohydrate\*frequency interaction on mean blood lactate concentration measured throughout the 30-km TTs ( $p > 0.05$ ; Table 2).

**\*\* INSERT FIGURE 2 AROUND HERE \*\***

## *Gastrointestinal discomfort*

Greater GI discomfort was apparent in HC-LF trials compared to all other trials. There was a treatment\*distance interaction effect for GI discomfort ( $p = 0.023$ , Figure 3). There was a significant effect of carbohydrate ingestion rate, frequency and a carbohydrate\*frequency interaction effect on mean GI discomfort ( $p \leq 0.05$ ; Table 2), with post-hoc analyses revealing a significantly higher mean GI discomfort in HC-LF compared with HC-HF ( $p = 0.002$ ), MC-LF ( $p = 0.020$ ) and MC-HF ( $p = 0.002$ ).

**\*\* INSERT FIGURE 3 AROUND HERE \*\***

## *Substrate oxidation and $\dot{V}O_2$*

Whole-body carbohydrate oxidation was decreased and lipid oxidation increased in LF compared to HF trials however there was no difference in  $\dot{V}O_2$  between trials. There was a significant effect of frequency ( $p \leq 0.05$ ) but no effect of carbohydrate ingestion rate or carbohydrate\*frequency interaction ( $p > 0.05$ ) on whole-body carbohydrate and lipid oxidation rates at  $1^\circ$  (Table 2). The LF trials elicited a greater reliance on carbohydrate oxidation (by  $\sim 6$ -7 %), and a reduction in lipid oxidation (by  $\sim 17$ -19 %) compared with the HF trials when matched for carbohydrate ingestion rate. There was no effect of carbohydrate ingestion rate, frequency or carbohydrate\*frequency interaction on mean  $\dot{V}O_2$  at  $1^\circ$  or  $7^\circ$  ( $p > 0.05$ , Table 2).

## *Race performance*

Performance times were  $142:41 \pm 17:12$ ,  $139:57 \pm 16:24$ ,  $140:27 \pm 17:11$  and  $137:35 \pm 15:24$  min:s in trials one to four, respectively, which reflected a significant effect of order ( $p < 0.001$ ). No order effect was apparent for any other variable ( $p > 0.05$ ). Performance times were  $140:11 \pm 15:31$ ,  $140:43 \pm 17:40$ ,  $139:12 \pm 15:32$  and  $140:33 \pm 17:46$  min:s in HC-HF, HC-LF, MC-HF and MC-LF, respectively (Figure 4). There was no effect of carbohydrate ingestion rate ( $p = 0.523$ ), frequency ( $p = 0.363$ ) or carbohydrate\*frequency interaction ( $p = 0.621$ ) on performance.

**\*\* INSERT FIGURE 4 AROUND HERE \*\***

#### *Heart rate and ratings of perceived exertion*

Mean heart rate was  $160 \pm 11$ ,  $160 \pm 9$ ,  $161 \pm 10$  and  $161 \pm 11$  beats·min<sup>-1</sup> at 1° and  $177 \pm 10$ ,  $176 \pm 7$ ,  $177 \pm 9$  and  $177 \pm 8$  beats·min<sup>-1</sup> at 7° in HC-HF, HC-LF, MC-HF and MC-LF, respectively. Mean RPE was  $16 \pm 1$  for each carbohydrate strategy. There was no effect of carbohydrate ingestion rate, frequency or carbohydrate\*frequency interaction on RPE or heart rate ( $p > 0.05$ ).

**\*\* INSERT TABLE 2 AROUND HERE \*\***

#### **Discussion**

The current study shows that doubling the rate of multiple-transportable carbohydrate ingestion from  $1.2 \text{ g} \cdot \text{min}^{-1}$  to  $2.4 \text{ g} \cdot \text{min}^{-1}$  of a 1:1 maltodextrin-fructose mixture did not affect gastrointestinal discomfort or metabolic responses during 30-km cross-country skiing TT

performance. However, ingesting this carbohydrate in two large feeds versus six smaller feeds resulted in poorer maintenance of euglycaemia, reduced lipid oxidation and increased GI discomfort, so may be less favourable for optimal exercise performance in general. Despite this, there were no effects of ingestion rate or feeding frequency on performance, although these findings must be considered in the context of a significant order effect for performance.

#### *Carbohydrate ingestion rate*

In the current study a variety of measures were taken to identify the potential effects of increasing the carbohydrate dose on metabolism and perceptions of GI discomfort. No order effect was apparent for any variable except performance time, thus the order effect for performance had no impact upon the characterisation of the GI discomfort and metabolic responses to the carbohydrate strategies. Gastrointestinal discomfort has been previously cited as a potential limiting factor on performance when ingesting high rates of carbohydrate (Newell et al., 2014). However, in the current study there was no difference between HC-HF and MC-HF, with GI discomfort peaking at a mean score of eight to nine (at 29 km), representing very light discomfort. This is in agreement with previous research in cycling (Jentjens et al., 2004; Jentjens et al., 2004; Triplett et al., 2010; Rowlands et al., 2012) and triathlon (McGawley et al., 2012), suggesting that high doses of multiple-transportable carbohydrates consumed little and often can be tolerated by the stomach without severe discomfort. Therefore, GI discomfort does not seem to be able to explain the discrepancy between high exogenous carbohydrate oxidation rates (Jentjens & Jeukendrup, 2005) and similar, or even diminishing (Smith et al., 2013), performance with increasing dose of multiple-transportable carbohydrate.



In terms of substrate availability, there was no effect of increasing carbohydrate dose above 1.2 g·min<sup>-1</sup> on blood glucose concentration, which is in agreement with previous studies (Jentjens et al., 2004; Jentjens & Jeukendrup, 2005). In contrast, blood lactate concentration is increased following the ingestion of fructose in combination with other carbohydrates (Jentjens et al., 2004; Jentjens et al., 2004; Jentjens & Jeukendrup, 2005; Baur et al., 2014), with lactate then available for oxidation in exercising muscle (Lecoultre et al., 2010). In the current study, increasing the dose of fructose ingestion had no effect on blood lactate concentration. However, the blood samples were taken after a demanding uphill section at 7°, where  $\dot{V}O_2$  was typically 88-90% of  $\dot{V}O_{2max}$ . This was associated with substantial lactate accumulation (with mean  $\pm$  SD values ranging between  $6.0 \pm 1.6$  and  $6.3 \pm 1.4$  mmol·L<sup>-1</sup>), which would likely mask any smaller differences that may be apparent due to the conversion of fructose to lactate (Lecoultre et al., 2010).

Previous studies have proposed a dose-response relationship between carbohydrate ingestion and endurance performance up to  $\sim 1.0$ - $1.3$  g·min<sup>-1</sup> (Mitchell et al., 1989; Smith et al., 2013), with similar (Baur et al., 2014) or diminishing performance thereafter (Smith et al., 2013). The current study is consistent with these suggestions, finding no difference between HC and MC. Despite this, it is possible that any performance effect may have been masked by the significant order effect. Specifically, when comparing between HC-HF and LC-HF every participant was quicker in the trial performed second out of these two particular trials.

**The lack of any improvement in performance when increasing the multiple-transportable carbohydrate dose from 1.2 to 2.4 g·min<sup>-1</sup> occurred despite previously observed elevated exogenous carbohydrate oxidation rates (of up to 1.75 g·min<sup>-1</sup>) exhibited at high ingestion rates (of up to 2.4 g·min<sup>-1</sup>) when using multiple-transportable**

**carbohydrates (Jentjens & Jeukendrup, 2005; Jeukendrup, 2010).** However, it may be speculated that the high exogenous carbohydrate oxidation rates reported with multiple-transportable carbohydrate ingestion above 1.2 g·min<sup>-1</sup> (Jentjens & Jeukendrup, 2005; Jeukendrup, 2010) overestimate the exogenous carbohydrate oxidation rates that can be attained at race intensities (i.e., 60-90% of  $\dot{V}O_{2max}$  in the current study). At higher exercise intensities (> 70% of  $\dot{V}O_{2max}$ ) than those used to calculate exogenous carbohydrate oxidation rates (~ 50-60% of  $\dot{V}O_{2max}$ ) gastric emptying (Costill & Saltin, 1974; Neufer et al., 1989) and intestinal absorption (Lang et al., 2006) may become limiting to carbohydrate delivery and therefore performance, such that it is similar to ingestion of a moderate amount of carbohydrate. Alternatively, ingestion of 1.2 g·min<sup>-1</sup> of carbohydrate may sufficiently spare endogenous glycogen stores, especially in non-fasted athletes, such that glycogen depletion does not become limiting over this duration of exercise (~ 2-3 h). It may be that greater exogenous carbohydrate oxidation from high-rate multiple-transportable carbohydrate ingestion only becomes important during ultra-endurance activities where, theoretically, ~ 1.2 g·min<sup>-1</sup> may not prevent glycogen depletion. Indeed, recent recommendations suggest the use of high rates of carbohydrate ingestion (~ 1.5 g·min<sup>-1</sup>) for endurance activities lasting longer than 2.5 hours (Jeukendrup, 2014). Further studies looking at ultra-endurance exercise are required to understand whether there are benefits of high-rate multiple-transportable carbohydrate ingestion (> 1.2 g·min<sup>-1</sup>) in certain race situations.

#### *Feeding frequency*

Drinking little and often has traditionally been promoted during sports performance and is typically applied in studies of endurance performance (Currell & Jeukendrup, 2008; Triplett et al., 2010; McGawley et al., 2012; Rowlands et al., 2012; Smith et al., 2013). Although intuitively attractive, this is mostly without direct supporting evidence of the efficacy over

other strategies, such as consuming large boluses infrequently. The effect of feeding frequency is particularly relevant to cross-country ski racing since the hands, specifically, are occupied with holding the ski poles and the upper body, in general, is important for generating propulsive forces (Hoffman & Clifford, 1992). Therefore, it may be of practical benefit for skiers to feed fewer times during a long-distance race in order to minimise the need to slow down to collect and consume a feed. However, in the current study LF drinking strategies resulted in unfavourable metabolic responses and increased gastrointestinal discomfort.

**An inability** to maintain pre-exercise blood glucose concentration throughout the TT was apparent in the LF trials in the current study. In the LF trials large peaks in blood glucose concentration were apparent following carbohydrate ingestion (i.e., at 4 and 19 km), which were followed by significant nadirs at 14 and 29 km (where no carbohydrate had been consumed for 14 km). Whilst the determinants of fatigue relating to substrate availability have not been examined in cross-country skiing, carbohydrate improves exercise capacity in cycling partly through maintenance of systemic glucose availability (Coyle et al., 1986). Therefore, an inability of the LF feeding strategy to maintain euglycaemia may contribute to the inability of some participants to maintain the **potential advantage of only having to slow down to drink for 90 s in the LF trials versus 150 s in the HF trials**. Moreover, reduced lipid oxidation during the LF compared with HF trials was apparent, which resulted in a greater reliance on carbohydrate oxidation. The reduction in lipid oxidation may be due to an elevated blood insulin concentration following the large carbohydrate feeds in the LF trials, which suppresses lipid mobilisation (Jeukendrup et al., 1999). Moreover, given that feeding frequency appears to have little effect on exogenous carbohydrate oxidation rates (Jeukendrup & Jentjens, 2000), the concomitant increase in whole-body carbohydrate oxidation and

reduction in lipid oxidation may suggest a greater reliance on endogenous carbohydrate stores and greater glycogen depletion in LF trials, which could be detrimental to performance (Tsintzas et al., 1996). Although the precise mechanisms of fatigue relating to substrate availability are unclear in cross-country skiing, LF drinking produces unfavourable metabolic responses in terms of blood glucose concentration and substrate oxidation, which in the absence of a favourable performance effect prevents advocating LF drinking strategies.

Increased GI discomfort was apparent in the LF drinking strategies, especially during HC-LF trials where the mean GI discomfort score peaked at 'somewhat heavy discomfort'. Due to limitations in the rate of carbohydrate absorption from the gut (Shi et al., 1995; Duchman et al., 1997; Jeukendrup et al., 1999), a large but diminishing proportion of the carbohydrate ingested in a single large bolus (~ 165 g of carbohydrate in the HC-LF trials) would remain in the gut for an extended period of time. This large but diminishing amount of CHO remaining in the gut could potentially have caused the transient pattern of GI discomfort apparent in the LF trials (Figure 4). Moreover, the degree of GI discomfort in the HC-LF trials, without accompanying improvements in performance compared to the HF trials, prevents the recommendation of this strategy.

Despite providing a potential 60 s performance advantage of not having to slow down to drink as frequently in the LF versus HF trials in the current study, there was no difference in performance between HF and LF strategies. However, any effect may have been masked by the order effect. Indeed, when comparing HC-HF with HC-LF, 12 out of 13 participants were quicker in the second of these particular trials and similarly, 12 out of 13 participants were quicker in the second trial when comparing MC-HF and MC-LF.

## *Limitations*

The order effect in the current study limits the confidence in the current findings relating to the **performance outcome**. There may be multiple reasons for the apparent order effect, including improving treadmill roller-skiing technique, improving pacing strategies with increasing familiarisation to the 30-km TT course (Swart et al., 2009) and rapid physiological adaption to the exercise stimulus (Hoffman & Clifford, 1992; Jeukendrup, 2010; Jeukendrup, 2014). While the extensive familiarisation strategy applied in the current study was designed to prevent improvements in treadmill roller-skiing technique throughout the trials, the possibility of this occurring cannot be excluded. Moreover, as the full 30-km TT was not completed prior to trial one this may have contributed to the apparent order effect, especially between trials one and two. However, maintaining motivation was also an important consideration and completing an additional full 30-km TT may also have led to a negative outcome.

## **Perspectives**

Recent studies have demonstrated a dose-response relationship between multiple-transportable carbohydrate ingestion and cycling performance, suggesting that cycling performance may peak at an ingestion rate of  $\sim 1.2\text{-}1.3 \text{ g}\cdot\text{min}^{-1}$  (Mitchell et al., 1989; Smith et al., 2013). However, the effect of carbohydrate ingestion during cross-country skiing has received very little research attention in the past and is relevant given the physiological differences between cross-country skiing and cycling or running (Bergström et al., 1973; Björklund et al., 2010; Losnegard et al., 2014). Moreover, this is the first study to analyse the interaction between carbohydrate dose and frequency. Neither doubling the rate of carbohydrate (1:1 maltodextrin-fructose) ingestion during exercise (from 1.2 to  $2.4 \text{ g}\cdot\text{min}^{-1}$ )

489 nor ingesting these doses more or less frequently affected performance of a 30-km cross-  
490 country skiing TT. From a practical perspective the less frequent feeding strategies (i.e., at 15-  
491 km vs 5-km intervals) may be inadvisable due to other potentially unfavourable responses,  
492 including poorer maintenance of euglycaemia, reduced lipid oxidation and increased GI  
493 discomfort.

494

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596 **TABLE LEGENDS**

597

598 **Table 1.** Contents of the two beverages provided during the high and moderate carbohydrate  
599 trials.

600

601 **Table 2.** Physiological responses during the four 30-km TTs.

## FIGURE LEGENDS

**Figure 1. A.** Schematic of the 30-km TT showing sampling of blood, RPE and GI discomfort (dashed arrows), GI discomfort only (dotted arrows), carbohydrate feeding (solid arrows) and expired gas collection periods (horizontal dashed arrows). W/U: warm-up. **B.** Profile of one lap of the 30-km TT course. Mean heart rate,  $\dot{V}O_2$  and  $\dot{V}CO_2$  were calculated for the final minute of the 1<sup>o</sup> and 7<sup>o</sup> sections.

**Figure 2.** Blood glucose concentration throughout the 30-km TT. \* denotes a significant difference between HC-HF and HC-LF; † denotes a significant difference between MC-HF and MC-LF.

**Figure 3.** Gastrointestinal discomfort throughout the 30-km TT. \* denotes a significant difference between HC-LF and HC-HF, † denotes a significant difference between HC-LF and MC-HF, ¥ denotes a significant difference between HC-LF and MC-LF.

**Figure 4.** Mean performance times (columns) with individual responses (lines). Trial order is indicated by marker shape: trial one (circle), trial two (diamond), trial three (triangle) and trial four (square).

621    **List of Supporting Information**

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